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RESEARCH ARTICLE

Uric acid–reducing efficacy of *Plectranthus amboinicus* (Lour.) Spreng in mice with potassium oxonate-induced hyperuricemia

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ABSTRACT

Plectranthus amboinicus (Lour.) Spreng is a type of Lamiaceae perennial herb that is widely used in folk treatments for respiratory and skin diseases. Notably, research on the hypouricemic effect of *P. amboinicus* is limited. This study investigated the uric acid–reducing efficacy of *P. amboinicus* in mice with potassium oxonate-induced hyperuricemia by performing an in vitro xanthine-oxidase-inhibition-activity-guided test of *P. amboinicus* extract (PAE) and its fractions. The ethyl acetate fraction (EAF) of PAE exhibited optimal efficacy in terms of the phenolic content, DPPH scavenging activity, and xanthine oxidase inhibition. We further demonstrated the uric acid–reducing activity of EAF in a mouse model of potassium bromate–induced hyperuricemia. The results can serve as a useful preclinical reference for researching the gout prevention effects of functional foods.

Keywords: *Plectranthus amboinicus* (Lour.) Spreng; Hyperuricemia; Uric acid–reducing activity

INTRODUCTION

Hyperuricemia is a common metabolic disease with harmful effects, and it is attracting increasing attention from researchers. Living standard improvements and lifestyle changes have caused the prevalence of hyperuricemia and gout to increase year by year. Hyperuricemia has become a prevalent chronic disease in various countries. The prevalence of hyperuricemia was estimated to be 10.6% in Thailand, 8.4% in Saudi Arabia, 6.4% in the middle-aged population of China, and 6.3% in Taiwan. In Taiwan, this prevalence is consistent because of poor gout management; notably, only one in five affected people undergo urate-lowering therapy (ULT) (Al-Arfaj, 2001; Kuo et al., 2015; Lohsoonthorn et al., 2006; Song et al., 2018; Wang et al. 2023). The prevalence of hyperuricemia and gout is increasing in the younger population, higher among men than among women, and higher in coastal areas than in inland areas. (Kuo et al., 2015; Song et al., 2018; Zheng et al., 2018; Teramura et al. 2023).

Clinicians are increasingly recognizing the harmful effects of hyperuricemia on various tissues and organs. Hyperuricemia is regarded as an independent risk factor for various metabolic diseases and cardiovascular conditions (e.g., metabolic syndrome, type 2 diabetes, hypertension, cardiovascular events and death, and chronic kidney disease). Because of the various harmful effects of elevated serum uric acid (SUA) levels, hyperuricemia is regarded as the fourth most crucial risk factor for chronic diseases after hypertension, hyperlipidemia, and diabetes. Metabolic and cardiovascular risk factors associated with elevated SUA should be actively managed with a treatment plan (Liu et al., 2018; Kou et al., 2021; Wang et al., 2022; Hu et al. 2023).

Two types of uric acid–reducing drugs are currently used in clinical settings, namely, drugs that inhibit uric acid synthesis (e.g., allopurinol) and drugs that increase uric acid excretion (e.g., benzbromarone and probenecid). The risk of severe hypersensitivity reactions is higher in the white population than in other ethnic or racial populations; when the

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etiological typing of hyperuricemia is unavailable, uricosuric drugs may be more effective than other drug options. However, uric acid–reducing drugs can cause various side effects; for example, allopurinol, benzbromarone, and febuxostat can cause Stevens–Johnson syndrome, cardiovascular disease, and hepatotoxicity (Pacher et al., 2006; Gerull et al., 2011; Balakirski & Merk, 2017; Lerch et al., 2018; Pascart & Lioté, 2019; Stack et al., 2021; Zeng et al. 2023). Because of the various risks of first-line drugs, developing therapies with fewer side effects is a key topic that can be explored by examining botanical resources (Wang et al., 2022).

Plectranthus amboinicus (Lour.) Spreng, a semisucculent perennial plant from the family Lamiaceae, is widely used in folk treatments for respiratory and skin diseases (Arumugam et al., 2016; Barbosa et al. 2023). *P. amboinicus* is also used as a substitute for oregano to mask the strong odors and flavors of fish, mutton, and goat (Staples & Kristiansen, 1999; Khan et al. 2023). *P. amboinicus* has been reported to have antioxidant, anti-inflammatory, antidiabetic, and antimicrobial properties (Arumugam et al., 2016; Barbosa et al. 2023; Sawant et al. 2023). However, few studies have examined the uric acid–reducing effect of *P. amboinicus*. In the present study, we demonstrated that *P. amboinicus* can reduce SUA concentrations in an animal model of hyperuricemia.

MATERIALS AND METHODS

P. amboinicus (Lour.) Spreng was purchased from Yuanshan Biotech Company, a local herb supplier based in Kaohsiung, Taiwan, in 2020. From Sigma-Aldrich (St. Louis, MO, USA), we purchased allopurinol, thymol, carvacrol, potassium oxonate (PO), Folin–Ciocalteu phenol reagent, xanthine, xanthine oxidase (XO), gallic acid, tocopherol, potassium dihydrogen phosphate, sodium dihydrogen phosphate, ethylenediaminetetraacetic acid, sodium chloride, potassium chloride, sodium carbonate, dimethyl sulfoxide (DMSO), and 1,1-diphenyl-2-picrylhydrazyl (DPPH). In addition, methyl alcohol, grain alcohol, acetone, phosphoric acid, and hydrochloric acid (HCl) were purchased from Echo Chemical (Miaoli, Taiwan). Other chemical agents, namely, *n*-hexane, ethyl acetate, and *n*-butanol, were obtained from Jiuh Hsing Instrument.

Preparation of *P. amboinicus* extract (PAE)

Fresh *P. amboinicus* was dried in the shade for 7 days and then smashed into granules. Granule powder was extracted with a 10-fold excess volume of 95% EtOH (w/v%) for 3 h and by using a reflux extraction apparatus (Angu, Kaohsiung, Taiwan). The extraction solution was filtered through paper in a funnel. PAE, which exhibits XO inhibitory activity, underwent liquid-phase extraction, during which *n*-hexane, ethyl acetate, *n*-butanol, and pure water fractions were used to obtain HxF, EAF, BuF, and WF, respectively. The solvents were removed by performing freeze-drying procedures; they were then stored in a drying cabinet at room temperature for subsequent experiments.

Analysis of total polyphenol content of PAE and its fractions

The total polyphenol content of PAE was estimated spectrophotometrically by causing a colorimetric redox reaction through the Folin–Ciocalteu reagent. In brief, 10 mg serial PAE was dissolved in 10 mL acetone solution (acetone: H_2O = 6:4). Subsequently, 0.2 mL PAE solution was mixed with 7.5% Na_2CO_3, and 1.0 mL Folin–Ciocalteu reagent was added to each well of a 96-well microplate (Jet Biofil, FEP-000-096, Guangzhou, China) and mixed thoroughly. After 30-min incubation at room temperature, the color change that occurred was measured with a spectrophotometer at 765 nm by using an enzyme-linked immunosorbent assay reader (BMG Labtech, SPECTROstar Nano, Ortenberg, Germany). The total polyphenol content was determined on the basis of micrograms of gallic acid equivalents (Wang et al., 2022).

Measurement of DPPH free radical scavenging activity of PAE

The antioxidant activity of PAE was measured using a DPPH free radical scavenging assay. PAE stock solution (1 mg/mL) was prepared and diluted with methanol to create sample solutions with different concentrations. The 50-μL aliquot of each dilution was added to a 96-well microplate. Subsequently, 150 μL DPPH working solution (250 μM) was added to each well; after 30 min, optical density was measured using a microplate reader at 490 nm. The half-maximal inhibitory concentration of PAE for scavenging DPPH free radicals was calculated and compared with that of vitamin E (Wang et al., 2022).

HPLC analysis of XO inhibition test

Uric acid samples of the PAE stock solution (1 mg/mL) were prepared and diluted with DMSO (DMSO: H_2O = 2:8) to attain various concentrations for performing an in vitro XO inhibition test. Subsequently, 250 μL test samples or samples with various concentrations were added to test tubes. Next, to cause a reaction, 175 μL of 50 mM phosphate buffer (pH 7.5) and 150 μL of XO solution (0.1 U/mL with 50 mM phosphate buffer, pH 7.5) were added at 25°C for 15 min, and 300 μL of 150 mM xanthine reagent (containing 50 mM phosphate buffer, pH 7.5) was added at 25°C for 30 min. Finally, 125 μL of 1 M HCl was added to stop the reaction. The reaction solution was filtered through a 0.45-μm filter for high-performance liquid chromatography (HPLC)–diode-array detection

(DAD) to monitor uric acid production. A Cosmosil $5C_{18}$-MS-II (4.6 × 250 mm^2, 5 μm) phase column was used; phosphate buffer: methanol (98:2) was used as the mobile phase, and a sample injection volume of 20 μL was added (flow rate of 1.0 mL/min for 15 min) to enable analysis at a wavelength of 280 nm. Allopurinol, an XO inhibitor, served as a positive control. Each sample was tested thrice, and the inhibition rate (%) was calculated using the following formula:

Inhibition of uric acid production (%) = (1 − ([Uric acid concentration in sample group]/[Uric acid concentration in control group]) × 100.

HPLC analysis of the indicated ingredient carvacrol and thymol in PAE and its fractions

Carvacrol, thymol, PAE, and fractions of the stock solution (1 mg/mL) were prepared and diluted with methanol to attain various concentrations for determining the composition of the samples. The samples were filtered through a 0.45-μm filter for HPLC-DAD to quantify carvacrol and thymol. A Cosmosil $5C_{18}$-MS-II (4.6 × 250 mm^2, 5 μm) phase column was used; acetonitrile: deionized water (60:40, v/v) was used as the mobile phase, and isolate elution (sample injection volume of 20 μL at a flow rate of 1.0 mL/min for 15 min) was performed to enable analysis at a wavelength of 220 nm.

Hyperuricemia animal model

In total, 48 male Institute of Cancer Research mice (ICR mice, male, 5 weeks, 25–27 g) were purchased from BioLASCO (Taipei, Taiwan) and housed under standard laboratory conditions in a temperature-controlled (22°C ± 2°C) animal facility with a 12-h/12-h light/dark cycle. The mice had ad libitum access to standard animal feed (LabDiet, LabDiet 5001, MO, USA) and sterilized water, and they were procured 1 week prior to testing to allow them to acclimatize to the laboratory environment and diet before the start of the experiments. The mice were randomly assigned to six groups each with six mice: the normal (N), vehicle control (VC), positive control (PC, 10 mg/kg allopurinol), low-dose (L, 125 mg/kg ethyl acetate fraction [EAF]), medium-dose (M, 250 mg/kg EAF), and high-dose (H, 500 mg/kg EAF) groups. For hyperuricemia induction, the mice in the VC, PC, L, M, and H groups were administered 250 mg/kg PO through subcutaneous injection every day for 7 days as per the protocol applied in other studies (Chen et al., 2019; Hu et al., 2010; Wang et al., 2022); after 1 h, the L, M, and H groups were administered 125, 250, and 500 mg/kg of EAF through oral gavage, respectively, and the PC group was administered 10 mg/kg allopurinol. After 1 week of drug administration, the mice were humanely sacrificed through carbon dioxide inhalation at the end of treatment, and their blood was immediately collected from the inferior vena cava into a tube with no anticoagulant to analyze serum concentrations (Murugan et al., 2019). The collected serum was analyzed for SUA, blood urea nitrogen (BUN), creatinine (CRE), glutamic oxaloacetic transaminase (GOT), and glutamic pyruvic transaminase (GPT) concentrations. This study was approved by the Animal Care and Use Committee of Tajen University (Pingtung, Taiwan; approval no. IACUC-108-15).

Statistical analysis

Data are expressed as means ± standard deviations, and each analysis was performed in triplicate. For statistical comparisons, SPSS version 16.0 (SPSS, Chicago, IL, USA) was used to conduct one-way analyses of variance and Tukey multiple comparison tests. A p value of <0.05 was regarded as statistically significant.

RESULTS

Polyphenolic compositions and free radical scavenging activity of PAE and its fractions

The polyphenolic compositions and free radical scavenging activity of PAE and its fractions were determined (see Figs. 1 and 2 for results). Our results indicate that ethyl acetate is the optimal solvent for extracting antioxidant polyphenols from PAE. The polyphenol and DPPH free radical scavenging activity of EAF were significantly higher ($p < 0.05$) than those of the other PAE fractions.

Xanthine oxidase inhibitory effect of PAE and its fractions

To screen for uric acid-lowering activity, the in vitro xanthine oxidase (XO) inhibitory activity of PAE and its fractions was evaluated (see Fig. 3 for results). The results revealed that at a concentration of 1000 μg/mL, the XO inhibitory activity of EAF was significantly higher than those of the other PAE fractions. Notably, the XO inhibitory effects of the ingredients carvacrol and thymol were substantially less pronounced relative to that of the PAE.

Effect of PAE on hyperuricemia in animals

We selected the EAF of PAE with the optimal XO inhibitory activity for further testing of hypouricemic effects in a hyperuricemic animal model. The hepatic and renal indices (i.e., CRE, BUN, GOT, and GPT) of the mice with PO-induced hyperuricemia did not reveal any significant hepatic or renal toxicity after EAF administration relative to the N group ($p > 0.05$; Table 1). Regarding SUA levels, EAF administration reduced PO-induced hyperuricemia in a dose-dependent manner. Therefore, PAE extracted with ethyl acetate is optimal for application as an uric acid–reducing treatment.

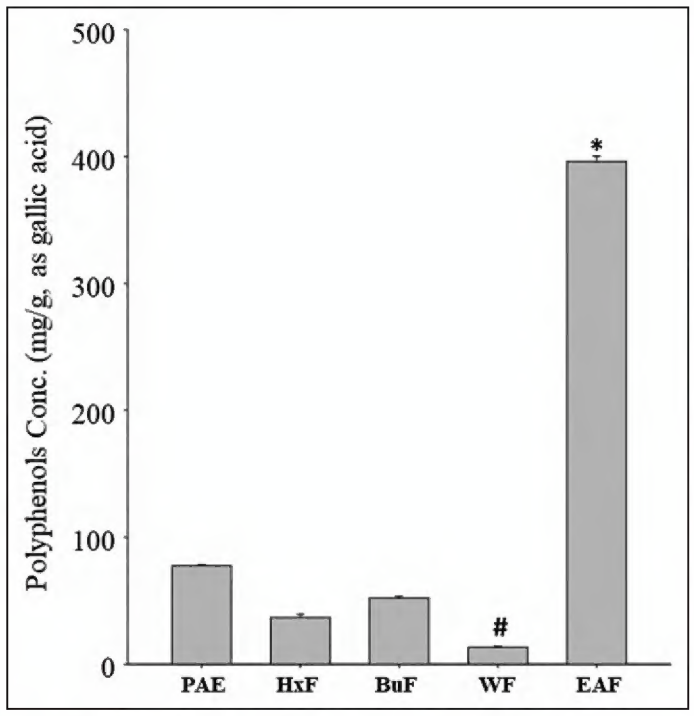

Fig 1. Total phenol content in *P. amboinicus* extract (PAE) and its fractions. Results are presented as means ± standard errors (n = 3). *p < 0.05 indicates significant increase relative to PAE, #p < 0.05 indicates significant reduction relative to PAE (Tukey's test). PAE, *P. amboinicus* extract; HxF, n-hexane fraction; BuF, *n*-butanol fraction; WF, water fraction; EAF, ethyl acetate fraction.

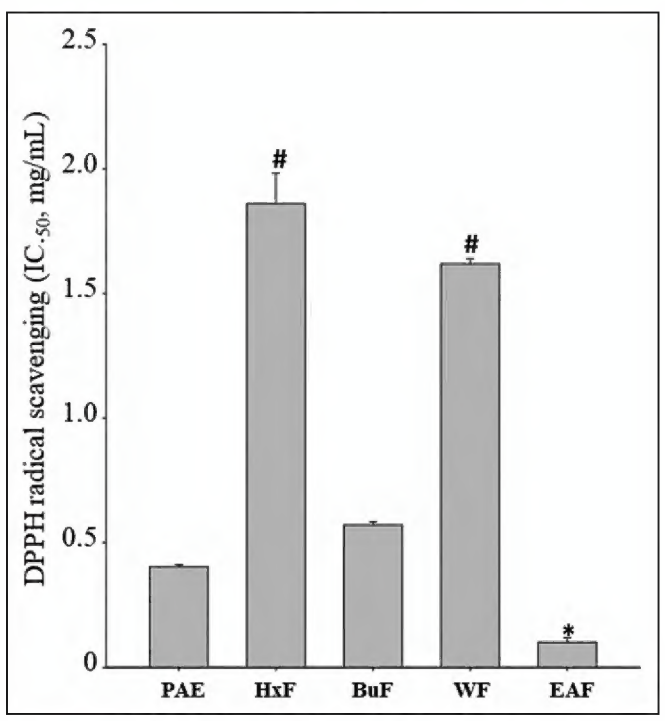

Fig 2. DPPH free radical scavenging activity of PAE and its fractions. Results are presented as means ± standard errors (n = 3). *p < 0.05 indicates significant reduction relative to PAE, #p < 0.05 indicates significant increase relative to PAE (Tukey's test). PAE, *P. amboinicus* extract; HxF, *n*-hexane fraction; BuF, *n*-butanol fraction; WF, water fraction; EAF, ethyl acetate fraction.

HPLC analysis of the indicated ingredients carvacrol and thymol in PAE and its fractions

The HPLC chromatograms of PAE and its fractions are provided in Fig. 4, and the carvacrol and thymol contents in PAE and its fractions are listed in Table 1. The retention time of the carvacrol and thymol peaks was 7.5 and 8.1 min, respectively. Carvacrol and thymol mainly exist in the *n*-hexane fraction (HxF), and the carvacrol content is higher than the thymol content. A comparison of the results of the XO inhibitory assay and HPLC revealed that carvacrol and thymol were not the major active ingredients contributing to the XO inhibitory effects of EAF.

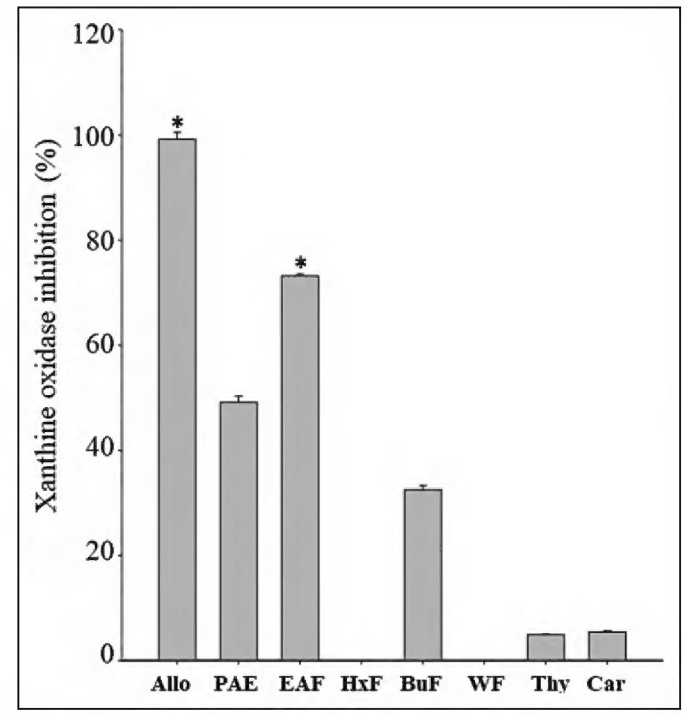

Fig 3. Xanthine oxidase inhibitory effects of PAE and its fractions (n = 3). Significant differences are determined using Tukey's test (*p < 0.05 for comparison with PAE). HxF, *n*-hexane fraction (1000 μg/mL); BuF, *n*-butanol fraction (1000 μg/mL); WF, water fraction (1000 μg/mL); EAF, ethyl acetate fraction (1000 μg/mL); Allo, allopurinol (100 μg/mL); Thy, thymol (1000 μg/mL); Car, carvacol (1000 μg/mL).

Table 1: Carvacrol and thymol contents in *P. amboinicus* extract and its fractions

Extracts and Fractions	Carvacrol (mg/g)	Thymol (mg/g)
PAE	7.46	0.95
HxF	43.49	3.57
EAF	5.72	1.85
WF	<0.1	<0.1
BuF	<0.1	<0.1

In vitro XO inhibitory effects of fractions of PAE

An analysis of the influence of the fractions of PAE on *in vitro* XO inhibition.

Results are presented as means ± standard errors (n = 6). *p < 0.05 versus normal group, #p < 0.05 versus VC group (Tukey's test). Six groups are normal (N), vehicle control (VC), positive control (PC), low-dose (L, EAF [150 mg/kg]), medium dose (M, EAF [250 mg/kg]), and high-dose (H, EAF [500 mg/kg]) groups. Abbreviations: CRE, creatinine; BUN, blood urea nitrogen; GOT, glutamic oxaloacetic transaminase; GPT, glutamic pyruvic transaminase; SUA, serum uric acid.

DISCUSSION

Gout and hyperuricemia are major public health problems, and hyperuricemia is a risk factor for gout. Allopurinol is the first-line medication for ULT (Jennings et al., 2014; Conley et al. 2023). Compared with other ethnic or racial populations, the Asian population is more susceptible

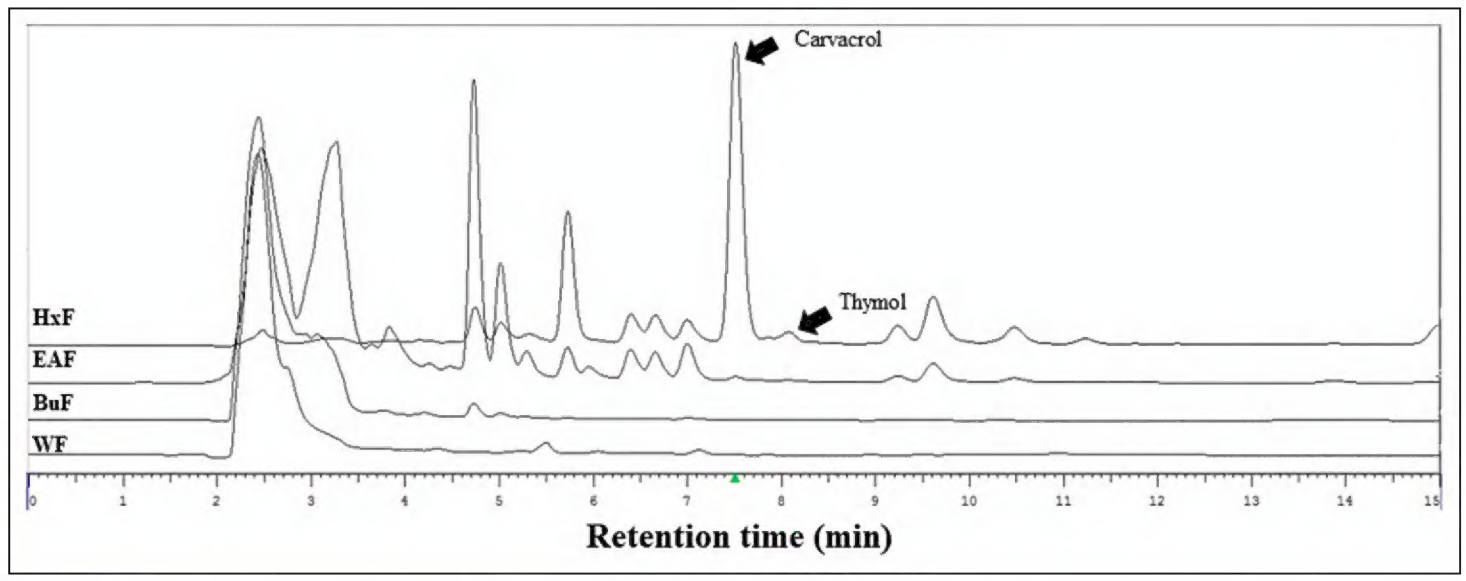

Fig 4. High-performance liquid chromatograms of PAE and its fractions. HxF, *n*-hexane fraction; BuF, *n*-butanol fraction; WF, water fraction; EAF, ethyl acetate fraction. Blue arrows indicate peaks of carvacrol and thymol.

to the side effects of the first-line drug allopurinol; this phenomenon is directly related to the positive rate of the leukocyte antigen HLA-B*5801 allele in the Asian population. Specifically, the positive rate of this gene is between 6% and 8% in the Chinese Han population and only 2% in the white population. Scholars have suggested that Asian individuals should undergo an HLA-B*5801 gene rapid PCR test prior to using allopurinol. In 2008, Taiwan introduced this gene test for patients with indications for allopurinol use, although it is not used for patients with positive results. With the public becoming increasingly aware of drug safety, the detection of this gene before medication use can reduce or eliminate the risk of adverse drug reactions (Somkrua et al., 2011; Ko et al., 2015; Tsukagoshi et al. 2023).

Gout attacks severely affect the work and quality of life of patients. The long-term treatment goals for patients with gout are to reduce their blood uric acid concentration, reduce their frequency of acute attacks, and improve their prognosis (Abhishek et al., 2016; Yutong et al. 2023). The low blood concentration of uric acid in gout patients is related to their poor compliance with uric acid–reducing therapy. The commonly used uric acid–reducing drugs of allopurinol, febuxostat, and benztropine are associated with low medication compliance because of their side effects. In Taiwan, only one in five patients with gout accept ULT. Only a small proportion of patients with gout exhibit high medication compliance, and those who do so visit the emergency room less frequently than those who do not (Becker et al., 2005; Richette & Bardin, 2010; Kuo et al., 2015; Ragab et al., 2017; Janssen et al., 2018; Wang et al., 2022).

For gout treatment, traditional Chinese medicine and Western medicine can be integrated to treat both the symptoms and the cause, and this strategy is drawing increasing attention from scholars (Kuo et al., 2017; Wang & Zhang, 2017; Liu et al. 2023). Research on adjunctive urate-lowering therapy has expanded; studies have evaluated the use of dietary ingredients to reduce the uric acid concentration and have demonstrated that the consumption of dogwood, lychee, and cherry (as a form of complementary therapy) is effective in reducing the uric acid concentration (Moriwaki et al., 2011; Zhang et al., 2012; Collins et al., 2019; Wang et al., 2022).

Studies have revealed that medicinal phytochemicals are highly complex and exhibit not only pharmacological effects (e.g., hypoglycemic, anti-inflammatory, antioxidant, and antitumor effects) but also an XO inhibitory effect; specifically, polyphenols can interact with amino acid residues at the active site of enzymes. It can change the secondary structure of XO, thereby inhibiting its activity; polyphenols and flavonoids are natural products that widely exist in various foods and plants (e.g., vegetables, fruits, and tea). Numerous flavonoids have been identified as potential XO inhibitors (Hatano et al., 1989; Moriwaki et al., 2011; Masuoka & Kubo, 2018; Chen et al., 2019; Xue et al. 2023).

In the present study, a similar trend was observed for PAEs with rich polyphenol and DPPH free radical scavenging activity (Fig. 1 and 2). Carvacrol and thymol are monoterpenoids found in *P. amboinicus*, and they have anti-inflammatory and analgesic effects. The current literature on *P. amboinicus* is mostly related to volatile essential oils; by contrast, in the present study, the identified *P. amboinicus* extract with optimal XO inhibitory activity is a nonvolatile oil compound. Plants from families such as the Lamiaceae, Rutaceae, Leguminosae, and Compositae families contain high levels of phenols, flavonoids, and tannins (Arumugam et al., 2016; Punet Kumar & Kumar, 2020). Polyphenols are used in herbal medicine for their antioxidant activity, and their antioxidants are regarded as herbal medicinal components. A main reason for the high efficacy of phenols is that it exhibits not only antioxidation properties but also antibacterial, anti-inflammatory, blood pressure–reducing, and anticancer properties. Overall, phenols have multiple physiological and pharmacological

Table 2: Biochemistry index for animal model

Groups	GOT	GPT	BUN	CRE	SUA
N	58.67±6.71	24.14±10.12	23.75±1.39	0.13±0.04	2.83±0.25
VC	156.33±10.07*	34.33±0.58	23.72±1.07	0.13±0.02	3.92±0.36*#
PC	120.33±11.02#	29.00±5.35	28.62±4.02	0.14±0.02	0.25±0.09#
L	57.00±7.18	22.67±3.79	24.43±2.99	0.12±0.03	3.36±0.27
M	66.75±11.73	27.80±8.35	25.00±2.39	0.11±0.03	2.92±0.27#
H	69.50±7.58	24.00±1.58	23.75±3.16	0.16±0.01	2.13±0.18#

effects that enable the scavenging of free radicals and the inhibition of oxidase activity, and they improve the activity of antioxidant enzymes. Phenolic molecules have large conjugated groups, and free radicals can easily react with the hydroxyl groups on the molecules of their hydrogen atoms to form phenolic-radical stabilized molecules, thereby delaying the oxidation chain reaction, reducing the destructive effects of free radicals, and inhibiting XO activity (Halliwell et al., 2005; Halliwell, 2008; Murthy et al., 2009; Fraga et al., 2010; Arumugam et al., 2016; Masuoka & Kubo, 2018; Mehmood et al., 2019; Rathod et al. 2023). In summary, thymol and carvacrol presented minor activity for XO inhibition (Fig. 3), and the HPLC chromatogram revealed other obvious peaks of EAF at 3.0–5.5 min and very small amounts of carvacrol and thymol (Fig. 3 and Table 1), indicating that the ingredients of carvacrol and thymol are not major active ingredients with XO inhibitory activity in EAF. Other active ingredients can contribute to the XO inhibitory activity of EAF.

Although hyperuricemia has no obvious clinical symptoms, elevated blood uric acid can activate the renin–angiotensin system, inhibit the synthesis of endogenous nitric oxide synthases, reduce the nitric oxide concentration, and promote the proliferation of arterial smooth muscle cells, which can lead to hypertension (Choi et al., 2014; Ubhadiya et al. 2023) and can promote the development of cardiovascular and renal diseases. Gouty nephropathy due to hyperuricemia has a similar pathogenesis. In addition to the deposition of uric acid crystals in renal arterioles, which can cause renal damage, gouty nephropathy can directly cause microvascular lesions in glomerular afferent arterioles, resulting in insufficient blood circulation in the kidneys and, consequently, chronic kidney disease (Johnson et al., 2013; Hu et al. 2023). A randomized controlled study that used allopurinol for treating hyperuricemia suggested that hyperuricemia is associated with renal impairment (Goicoechea et al., 2010; Bakhshaei et al. 2023); notably, few studies have explored the antihyperuricemic property and mechanism of allopurinol. In addition, Chinese herbal antigout drugs still mainly target XO and renal uric acid rotation, and few studies have explored other targets. In the present study, a mouse model of acute hyperuricemia induced by PO was established to assess the effect of plant on SUA levels in the mice, clarify the mechanism of this effect, and facilitate the development of alternative antigout therapy (Chi et al., 2020; Pu et al., 2021).

Among PAF and its fractions, EAF exhibited optimal XO inhibitory activity, as revealed by in vitro XO inhibitory activity-guided screening experiments. The uric acid–reducing efficacy of EAF in mice with PO-induced hyperuricemia was assessed. Table 2 presents that the SUA concentration in the VC group was significantly higher than that in the N group ($p < 0.05$). At an allopurinol dose of 10 mg/kg, the PC group exhibited a significantly reduced SUA concentration relative to that in the model group mice ($p < 0.05$). The mice with hyperuricemia in the M and H groups exhibited significantly reduced SUA concentrations ($p < 0.05$). Notably, EAF reduced the SUA concentration to approximately normal levels, whereas PC reduced the uric acid concentration to a less-than-normal level. Finally, the uric acid–reducing activity of EAF was validated in the mice with PO-induced hyperuricemia.

CONCLUSION

The EAF of PAE exhibited optimal efficacy in terms of the phenolic content, DPPH scavenging activity, and XO inhibition; we also validated its hypouricemic efficacy in mice with PO-induced hyperuricemia. PAE with ethyl acetate is optimal for future application as an alternative uric acid–reducing treatment. The results of this study contribute to the development and use of health-care products and alternative medicines for treating hyperuricemia and gout.

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CONFLICT OF INTEREST

The authors have no conflicts of interest to declare.

Author's contributions

Chih-Chiang Wang, Chun Chen and Fu-An Chen designed experiment and methodology. Chun Chen, Chih-Wei Chang and Po-Yen Chiu performed experiment. Chih-Chiang Wang, Chun Chen, Chih-Wei Chang and Po-Yen Chiu performed data curation. Chih-Chiang Wang, Chih-Wei Chang, Po-Yen Chiu and Fu-An Chen performed formal analysis. Chih-Chiang Wang and Fu-An Chen were supervising the research project. Chih-Chiang Wang, Chun Chen and Fu-An Chen prepared the original draft. Chih-Chiang Wang, Chun Chen and Fu-An Chen were involved in writing, review and editing.

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